proteins, but causes the formation of misoriented and non-polar hair bundles, indicating a role for ciliary genes in intrinsic cell polarization downstream of asymmetric PCP complexes. We further found that basal body positioning correlates with the polarity or loss of polarity of hair bundles and that basal body configuration appears to be affected in ciliary mutants. Strikingly, similar defects in the basal body and in the loss of intrinsic polarity were found in mouse mutants with defective Usher genes Usher proteins make up the machinery for the formation of polarized hair bundles. We have also detected a genetic interaction between Usher gene PCDH15 and ciliary gene IFT88 in basal body location and hair bundle morphogenesis. Together, our results suggest that ciliary genes act with Usher genes to configure the basal body to direct the formation of intrinsically polarized hair bundles.

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Program/Abstract # 214
Wnt/planar cell polarity signaling controls endoderm cell rearrangements during the morphogenesis of the primitive gut tube

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To generate normal functional anatomy in the digestive tract, the primitive gut tube (PGT) must undergo dramatic elongation and form a lumen lined by a single layer of polarized digestive epithelium. In *Xenopus* embryos, endoderm cells in the core of the PGT radially intercalate during gut elongation, but the morphogenetic mechanisms underlying these rearrangements are unknown. We previously showed that inhibition of Rho/ROCK/Myosin II activity prevents endoderm intercalation and consequently perturbs both gut elongation and digestive epithelial morphogenesis. Here we show that gut morphogenesis is governed by Wnt/PCP signaling. Gut-targeted expression of a dominant negative form of *Wnt11*, or an allele of *Disheveled* (*Dsh*) that specifically inhibits noncanonical Wnt signaling, results in shortened and malrotated gut tubes. Wnt11- or Dsh-deficient endoderm cells lose their polarized morphology and fail to properly intercalate. Moreover, exposure of late stage embryos to small molecule inhibitors of Rac or JNK perturbs the normal cell shape and adhesion patterns necessary for endoderm intercalation, and consequently induces severe defects in gut elongation and digestive epithelial morphogenesis. Our results suggest that the morphogenetic events driving tissue elongation in the PGT are mechanistically analogous to those that function during gastrulation. We propose that different Wnt/PCP signaling components control distinct endoderm cell properties and behaviors to coordinate the development of an epithelial lining with tubular tissue elongation in the PGT.

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Program/Abstract # 215
Wnt5b/Ryk signaling mediates polarized cell movement in zebrafish

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The Wnt signaling network plays an important role in patterning and morphogenesis. Wnt pathways via the seven-transmembrane receptor Frizzled (Fzd) regulate convergent extension (CE) movement in vertebrate embryos. Wnt has also been shown to signal through Ryk, an atypical receptor tyrosine kinase, to mediate axon guidance. However, the molecular mechanism of Wnt/Ryk signaling and its role outside the nervous system are less well characterized. Here we report a role of Wnt5b/Ryk signaling in zebrafish gastrulation. We combined gene knockdown and transplantation assays to show that Wnt5b/Ryk signaling is required for the CE movement during zebrafish gastrulation. We further demonstrate that Ryk internalizes into caveolin-coated endocytic vesicles upon Wnt5b stimulation and promotes polarized filapodia in migrating cells. While Wnt5b signaling through Ryk is independent of nuclear beta-catenin function, Ryk deficiency partially blocks Wnt5b-induced Disheveled (Dvl) turnover and Ryk overexpression activates intracellular calcium release, suggesting that Wnt5b/Ryk signaling regulates polarity effectors in common noncanonical Wnt pathways. In contrast to its role as a permissive cue in Wnt/Fzd signaling, Wnt5b transduces directional signals to Ryk-expressing cells. Our findings indicate that non-canonical Wnt ligands can modulate polarized cell movement in vertebrates by two mechanisms: a known mechanism by which activation of the core components of planar cell polarity (PCP) pathway through Fzd leads to establishment of polarity framework; and a novel mechanism by which Ryk signaling provides directional information for cell migration.

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Program/Abstract # 216
The PCP effector Fritz governs microtubule assembly and ciliogenesis in vertebrate multi-ciliated cells

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Cilia are microtubule-based organelles protruding from nearly all vertebrate cells. Several core components of the PCP signaling are essential for ciliogenesis. Fritz is an effector in the PCP signaling. Here, we examined its function in *Xenopus laevis* using antisense morpholino-oligonucleotides (MOs). Confocal microscopy and scanning electron microscopy revealed that axonemes on multi-ciliated cells of Fritz morphants were far shorter and fewer in number, as compared to controls. We observed that loss of Fritz results in the accumulation of apical cytoplasmic microtubules including polyglutamylated tubulins. Polyglutamylation is important for cilia assembly and function. A dramatic increase in polyglutamylated tubulin signal in the apical cytoplasm of Fritz morphant multi-ciliated cells indicates that the ectopic microtubule assembly in Fritz morphant is highly glutamylated. Next, we identified the CCT as an interacting partner of Fritz. CCT is a chaperonin and has been implicated in ciliogenesis. We generated GFP- or myc-tagged CCT subunit constructs and found that the GFP- or myc-tagged CCTa and CCTe were localized in punctate structures along the ciliary axonemes of multi-ciliated cells. We observed that loss of Fritz results in the accumulation of CCT at the apical cytoplasm in multi-ciliated cells. We suggest that by deregulating the localization or function of CCT in Fritz morphants, the turnover of microtubules may slow in the apical cytoplasm of multi-ciliated cells. As a result of reduced turnover, these microtubules may be longer-lived and thus acquire a higher concentration of polyglutamylation.

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Program/Abstract # 217
Specific cellular behaviors regulate in LR asymmetric heart looping

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Establishment of leftright (LR) asymmetry is required for correct positioning and function of internal organs. Our long-term goal is to reveal the mechanism of how LR asymmetric signals control LR asymmetric morphogenesis. We have reported that the initial C-looping process of fusing heart is regulated by two independent morphological events: the outflow tract rotation to the right side and overriding of the left caudal rudiments to the right side (Kidokoro et al. Dev Dyn 2008). Now we are focusing on LR differences at cellular level. During the C-looping, the left caudal rudiment becomes bigger than the right one. We found that the difference was not caused by increased cell numbers but cell shape changes. Similar changes were observed in the rostral region of looping heart tubes, suggesting that there is common molecular/cellular basis to generate leftright asymmetry between the rostral and caudal regions of the C-looping heart. Based on these data we will discuss how LR signals regulate cellular mechanisms such as cytoskeletal rearrangement and cell adhesion in heart looping, and how the common cellular machinery can control the different LR asymmetrical morphogenesis in the two regions of the C-looping heart tube. We will also discuss how the chick and mouse have similar and different morphogenesis during heart looping.

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Program/Abstract # 218
Identification of genes required for axis elongation in *Drosophila*
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During axis elongation in the *Drosophila* embryo, polarized cell movements cause the embryo to more than double in length along the anteriorposterior axis while narrowing in width along the dorsalventral axis. These cell rearrangements are accompanied by an asymmetric localization of actomyosin cables and adherens junction proteins that are required for polarized cell behavior. An oriented actomyosin network could provide the spatial information that guides cell rearrangement, while differential adhesion may influence local interactions between cells. We are carrying out a germline clone screen to identify additional genes required for axis elongation. This screen makes use of a collection of insertion mutations generated by Liqun Luo's lab at Stanford (Schuldiner et al., 2008) to produce mosaic females whose progeny are maternally deficient for specific genes. To date we have screened 627 lines, each with a unique, molecularly mapped insertion. Of these, 26 are required for embryonic patterning, 20 are required for early syncytial development, and 60 fail to complete axis elongation and have been selected for further analysis. Genes required for elongation could establish planar cell polarity or, alternatively, act at a later step to translate these polarities into directional cell behavior. To distinguish between these possibilities, we are performing secondary screens to ask if cytoskeletal and junctional proteins are asymmetrically localized in mutant embryos and live imaging analysis to associate these genes with specific aspects of polarized cell behavior.

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Program/Abstract # 219
Ectoderm membrane rafts regulate constituent nitric oxide synthases for myotome formation in chicken embryos
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Signaling molecules like nitric oxide (NO) and intracellular free Ca2+ are essential for embryonic growth. In this regard, multifunctional membrane rafts (rafts) have constitutive nitric oxide synthases (NOS) that require Ca2+-calmodulin binding for NO formation. Rafts are abundant structures in ectoderm, somites and neural tube of chicken embryos and are important to somite myotome formation as shown by loss of ectoderm rafts with MBC or by L-NAME blocked NOS activity. We now report dynamic NO signaling from ectoderm to somites and segmental plate mesoderm in embryos using DAF2-DA, a NO specific probe, and confocal microscopy. We show that ectoderm initiates a dynamic increase in NO (20min) to saturation before activating a second dynamic rise in NO (25min) in the anterior SPM and in early somites (ssI, ssVII). In ectoderm, NO increases in a few cells in the outer periderm layer before elevating globally across the ectoderm. A second NO rise follows in the dorsal epithelium of SPM and somites, but not in mesenchymal tissues. In new somites (ssI), the NO rise is weak and radiates across its dorsal surface. In contrast, the NO rise in myotome-forming somites (ssVII) exhibits an initial rise in the central dermomyotome (non-lip) that increases to saturation before transitioning medio-laterally into the dermomyotome dorsomedial lip. Furthermore, MBC or L-NAME treatment of the ectoderm blocks NO increases in the ectoderm and in both SPM and somites. We conclude that ectoderm rafts regulate constitutive NOS isoforms for NO signaling to somites for myotome formation.

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Program/Abstract # 220
The role of stromal derived factor-1α in vertebrate somitogenesis
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Somitogenesis is an important embryological process as it establishes the segmented nature of the vertebrate body. Previous work in zebrafish identified the cytokine, stromal derived factor-1α (Sdf-1α) as playing an important role in somite cell rotation (Hollway et al., 2007). Here we show that Sdf-1α is also important for somite morphogenesis in the frog, *Xenopus laevis*. We use a morpholino approach to knock down both Sdf-1α and its receptor, CXCR4. We show that Sdf-1α and CXCR4 morphant embryos do not undergo proper segmentation and somite rotation. These embryos fail to form properly aligned myotome fibers. Furthermore, MyoD and 12/101 expression levels are reduced in morphants in comparison to control embryos. Previous research using cell lines identified RhoA and Rac1, two small GTPases involved in actin regulation, as downstream targets of the SDF-1 signaling pathway (Bartolome et al., 2004; Tan et al., 2006). Based on these studies, we examined the effects of knocking down RhoA and Rac1 function. Confocal images of these embryos revealed a similar phenotype to that of Sdf-1α and CXCR4 morphants. We propose that the Sdf-1α signaling pathway may act through the activation of actin regulatory proteins, Rho A and Rac1, to regulate the changes in cell behavior necessary for the formation of properly aligned myotome fibers. Furthermore, this pathway appears to be conserved among vertebrates.

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Program/Abstract # 221
Zebrafish poky/CHUK/IKK1 is required for epiboly and EVL differentiation
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Zebrafish epiboly requires coordinated rearrangements of three distinct cell layers. The deep layer, enveloping layer (EVL) and yolk cell